Xanthan gum production using Xanthomonas campestris B6720: fermentation process and application in fermented soymilk

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Abstract. The production of xanthan gum using *Xanthomonas campestris* B6720 was investigated. The fermentation process, encompassing substrate consumption, bacteria growth, gum yield, explored and compared the produced gum with a commercially gum using FTIR. The study also explored application of xanthan gum in fermented soymilk. Investigating the addition of xanthan gum to fermented soymilk aims to assess its impact on sensory attributes, microbial stability, and overall stability of the product during storage. About 8.837 ±0.199 g/L of xanthan gum was obtained and 7.093 ±0.267 g/L of reducing sugars residuals at the end of the fermentation period. FTIR results revealed the similarities between the gum produced and the commercial gum. The inclusion of 0.020% of xanthan gum could have a positive effect on the physicochemical and microbial stability of fermented soymilk during storage and hence increasing consumer acceptability. The findings from this research hold promise for optimizing the production of xanthan gum using *X. campestris* B6720 and offer insights into its potential application in enhancing the sensory attributes and stability of fermented soymilk. This could have significant implications for the food industry, providing a valuable avenue for the utilization of xanthan gum as a functional ingredient in dairy alternative products.

1 Introduction

Xanthan gum, an adaptable biopolymer, has gained considerable attention across multiple industries due to its distinctive rheological attributes, stability, and compatibility with diverse substrates. Produced by the bacterium *Xanthomonas campestris*, this gum has seen widespread use in the food, pharmaceutical, and other industrial domains due to its capabilities in thickening, stabilizing, and forming gels and its natural antioxidant activity [1,2]. Its cost-effectiveness stands out in comparison to other microbial polysaccharides, given its high efficacy even in small amounts. Furthermore, it retains its rheological characteristics consistently across a wide spectrum of temperatures and pH levels in food products [3]. The gum's rheological properties can also be influenced by factors such as the carbon source, production conditions, and bacterial strain employed in its production.

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The trend toward adopting dairy-free diets has seen a significant surge in recent years, with non-dairy milk alternatives gaining substantial popularity [4]. This shift is primarily attributed to various factors, including milk allergies, lactose intolerance, concerns about high cholesterol levels, and the embrace of vegan diets [5]. Among these alternatives, soymilk derived from soybeans stands out as one of the most widely consumed and preferred plant-based milks globally, boasting a notably superior nutritional profile compared to other alternatives [6]. Fermented soymilk, a derivative of soymilk, holds particular promise due to its potential to positively impact the digestive gut environment and influence the composition of the gut microbiome. This aspect has sparked interest in exploring its role in anti-aging processes by regulating the gut microbiome, as emerging research suggests a strong link between gut health and the aging process [7]. One of the primary benefits of fermenting soymilk lies in its ability to mitigate the inherent beany flavor associated with soy products [8], thereby enhancing its palatability. Additionally, the fermentation process enriches soymilk with functional attributes, potentially amplifying its beneficial effects on health beyond its inherent nutritional value.

Vegetable milk-derived products lack consistency and texture despite their high consumer demand [9]. These products exhibit diminished colloidal stability, making them susceptible to sedimentation, separation, or syneresis during storage [10]. These occurrences lead to undesirable phase separation, compromising the overall product quality. To enhance the quality of such food items and ensure consumer acceptance while extending their shelf life, the incorporation of polysaccharide-based stabilizers becomes imperative. These stabilizers play a crucial role by effectively preventing ingredient separation, acting as emulsifiers and thickeners [10-12]. Therefore, the utilization of xanthan gum in fermented soymilk presents an intriguing avenue due to its ability to improve mouthfeel, prevent syneresis, and modify the final product thereby increasing the consumer acceptability. However, at present there is not enough research on this problem [17].

This research aims to delve into the production of xanthan gum using *X. campestris* B6720, exploring the fermentation process including substrate consumption, bacteria growth, gum yield, and the gum's conformity with the commercial gum. Additionally, the study also focuses on application of xanthan gum in fermented soymilk, examining its potential to enhance the sensory attributes, microbial stability, and stability of this dairy alternative on storage.

2 Materials and methods

2.1 Materials

Soybean was purchased from BKR Barnaul, Lenina Street 54, Yekaterinburg. The commercial xanthan gum was purchased from IP Nimchenko V. V., Krasnogo Mayaka street 16B, Moscow. The starter culture for the fermentation of soymilk (*Streptococcus thermophilus* and *Lactobacillus bulgaricus*) was also purchased from Kapria, Moscow (www.skvaska.com). Sweetener purchased from Solvi Rus LLC, 195027, St Petersburg, Russia. The vanilla flavor was purchased from REC-HAAS LLC, 105082, Moscow, Russia. The bacteria, *X. campestris* B6720 strain (lyophilized) was purchased from Bioresources Center All-Russian Collection of Industrial Microorganism (BRC VKPM), National Research Center (Moscow, Russia).

2.2 Methods

2.2.1 Biosynthesis of xanthan gum

The lyophilized *X. campestris* cells were rehydrated for 15 minutes in malt extract (1.7 g), peptone (1.0 g), and sterilized distilled water (100 ml) before further inoculation. About 0.1 ml aliquot of the rehydrated cell mixture was plated on a glucose-yeast extract agar plate (glucose 2 g, yeast extract 1 g, peptone 1 g agar 1.7 g, and distilled water 100 ml). The culture was incubated at 28 ^{O}C for 48 hours.

The nutrient media for biosynthesis was prepared containing 2.0 g Glucose, 0.3 g Yeast extract, 0.01 g Magnesium Sulphate ($MgSO_4.7H_2O$), 0.2 g Potassium Hydrogen Phosphate (K_2HPO_4), and 0.2 g Potassium dihydrogen Phosphate (KH_2PO_4) in 100 ml of distilled water. A loop of the bacteria culture from the agar plate were dissolved in a 10 ml in two different conical flasks containing the nutrient media. The cell culture was incubated at 28 ^{O}C for 24 hours at 250 rpm. After the 24-hour period, 5 ml of the pre-inoculated biosynthesis media was transferred into a 250 ml conical flask containing 100 ml of biosynthesis medium with the same composition as the pre-inoculation media. The resulting content was incubated on a shaker 250 rpm at 28 ^{O}C for 72 hours using orbital sharker-incubator (Biosan ES-20, Latvia). The optical density of the fermentation broth, gum yield, and glucose depletion in the media were determined and recorded every 24 hours. The experiment was conducted in triplicates.

2.2.2 Optical density measurement

The optical density (OD) of each sample in different times (0, 24, 48, and 72 h) was determined and recorded using Shimadzu UV-1800 spectrophotometer (Shimadzu, Japan) at λ =600 nm.

2.2.3 Total reducing sugar determination

The total soluble sugar content was determined with phenol–sulfuric acid method as described by Nielsen [13]. The reading was made at a wavelength of 487 nm using a glucose solution as the standard at concentrations of 10–50 mg/L.

2.2.4 Xanthan gum yield determination

About 10 ml of the broths were taken every 24 hours for analysis. The broth was centrifuged at 5000 rpm for 40 min to remove cells (biomass) and the gum contained in the supernatant was precipitated using isopropanol in the ratio 1: 3. The precipitate (gum) was dried at 50 ^{O}C for 48 hours in a hot air oven and the weight obtained. The gum was then milled using mortar and pestle and characterized using infrared spectroscopic analysis.

2.2.5 FTIR of xanthan gum

The gum was characterized using IR spectroscopy for both the commercial and the gum produced using *X. campestris B6720*. Infrared spectra were obtained using BRUKER alpha Fourier-transform infrared (FTIR) spectrometer, (Bruker, Germany) at room temperature approximately 22 ^{O}C.

2.2.6 Soymilk extraction and fermented soymilk preparation

Soybeans were weighed (120 g) and soaked in distilled water overnight at room temperature (in the ratio 1:10). The water was drained, and the soybean blended until a fine slurry was obtained and then strained to extract the soymilk and the residue, okara discarded (about 1.2 L of water was added during the blending). The soymilk was boiled for about 15-20 minutes. After cooling to a temperature of about 80 ^{O}C, 4 g of table salt, 0.3 g of vanilla flavor, and 10 g of sweetener (erythritol, stevia leaf extract, stevioside) were added to 1 liter of the milk. The soymilk was again pasteurized at 85 ^{O}C for about 5 minutes and a 100 ml were measured into glass jars and capped. All jars were treated with xanthan gum solutions (0.000% (control), 0.005% (Treatment 1, T1), 0.010% (Treatment 2, T2), 0.015% (Treatment 3, T3), and 0.020% (Treatment 4, T4)). About 0.1 g of the starter culture (*Streptococcus thermophilus and Lactobacillus bulgaricus*) was added to each sample on cooling to about 40 ^{O}C and stirred uniformly to allow even distribution. The samples were closed airtight and incubated at 42 ^{O}C for 6 hours. The samples were then stored in a refrigerator at 4 ^{O}C for further use.

2.2.7 Measurement of syneresis and water holding capacity (WHC)

The syneresis was determined using the method described in the work of Raikos et al. [9]. About 5 g of the samples were weighed onto a 2-V folded filter paper (Whatman) and placed on the top of a funnel. Syneresis was determined by gravity by measuring the weight (g) of liquid collected in a measuring cylinder of known weight. The drainage time was about 120 minutes and was done at room temperature (appr. 22 ^{O}C). The percentage of syneresis was calculated according to the equation:

$$Syneresis\ (\%) = (Weight\ of\ whey / Weight\ of\ sample)*100 \quad (1)$$

WHC of the samples were determine by centrifugation of about 5 g of the samples at 4500 rpm at 4 ^{O}C for 30 minutes. The water suspended was removed and weighed. The WHC was calculated as a percentage using the equation:

$$WHC\ (\%) = 1-(Weight\ of\ whey / Weight\ of\ sample)*100 \quad (2)$$

2.2.8 pH measurement

The pH of the samples was measured at 4 ^{O}C using FiveEasy pH meter (Mettler Toledo Ltd., Victoria, Australia).

2.2.9 Lactic acid bacteria count

Colony-forming units (CFUs) of bacteria was counted using a plate-counting method. Fermented soymilk samples (1 mL) were dispersed in 9 mL of sterile water to achieve a 10-fold dilution. A 0.1 mL aliquot from each serial dilution (10^{7}) was spread over an agar plate with de Man, Rogosa, and Sharpe (MRS) for lactic acid bacteria, LAB. The Petri dishes were incubated upside down at 37 ^{O}C for 48 hours and the colonies of the bacteria counted and reported as Log of colony forming units/mL (logCFU mL $^{-1}$).

2.2.10 Sensory analysis

Sensory analysis for the fermented soymilk blended with xanthan gum was performed within 24 hours after production and storage at a temperature of 4 ^{O}C by untrained panels (staff and graduate students at the Institute of Chemical Engineering, Ural Federal University,

Yekaterinburg, Russia). Evaluating flavor, appearance, taste, and overall acceptability on a 5-point hedonic scale (1-dislike extremely, 2-dislike, 3-neither like nor dislike, 4-like, 5-like extremely) for each sample. Water was used as a palate cleanser in between samples by the panellists.

2.2.11 Statistical analysis

All statistical analyses were executed using GraphPad Prism 8 statistical software package. The data obtained were analyzed using one-way analysis of variance (ANOVA) and the Tukey's multiple comparison test was used to compare means. The sensory data were analyzed using Two-way ANOVA with Dunnett's multiple comparisons test.

3 Results and discussions

3.1 Optical density

Monitoring the optical density (OD) of the biosynthesis medium provides insights into bacterial growth and product formation. The OD is often used as a proxy for biomass concentration in microbial cultures. A higher optical density usually corresponds to increased cell density. Figure 1 shows the OD of the medium during xanthan gum biosynthesis. According to the graph, there is a significant ($p < 0.0001$) increase in optical density from 0.096 ±0.001 to 1.045 ±0.060 within the first day. This rapid increase suggests robust bacterial growth and initial xanthan gum production. The optical density continues to increase significantly ($p < 0.0010$) from 1.045 ±0.060 to 1.277 ±0.048 on Day 2. This signifies that bacterial growth and xanthan gum production are still ongoing, although the rate might have slowed down compared to the initial phase. Between Day 2 and the final day, there is either a stabilization or a slight decrease in the optical density from 1.277 ±0.048 to 0.956 ±0.047. This could indicate several possibilities. For instance, the growth might have reached a plateau where the bacteria entered a stationary phase, leading to the stabilization of biomass. Factors such as nutrient depletion, accumulation of metabolic by-products, or limitations in the culture environment might have affected bacterial growth and consequently, xanthan gum production, leading to a decline in optical density. Alternatively, it is possible that some bacteria may have started to lyse or die off, reducing the overall biomass and hence, the optical density.

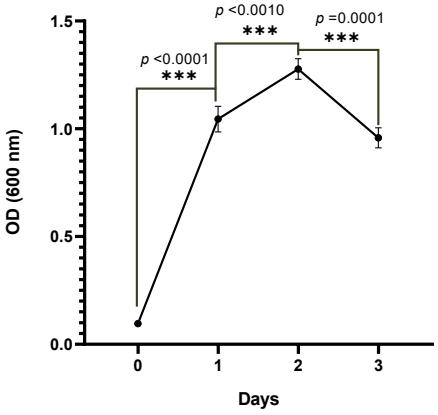

Fig. 1. Optical density of medium. Points are means (n= 3) and error bars are standard deviation of means, where *** are the significant differences.

3.2 Total reducing sugars content

The measurement of total reducing sugars content in the medium during the xanthan gum production experiment provides additional insights into the utilization of sugars by the bacteria for growth and the synthesis of xanthan gum. There is a significant ($p < 0.0001$) decrease in total reducing sugars content of the fermentation broth from 21.609 ±1.199 g/L to 13.317 ±1.628 g/L on Day 1. This reduction suggests that the bacteria are actively utilizing the available sugars for growth and metabolic processes, including xanthan gum production. The decreasing trend in total reducing sugars continues from 13.317 ±1.628 g/L to 12.357 ±0.400 g/L on Day 2. This indicates that the bacteria are still utilizing the sugars present in the medium for their growth and metabolic activities. There is a continued decrease significantly ($p < 0.0012$) in total reducing sugars from 12.357 ±0.400 g/L to 7.093 ±0.267 g/L by the final day of the experiment. This ongoing reduction as shown in figure 2 implies sustained bacterial activity and utilization of the available sugars, although at a slower rate compared to the initial stages of the experiment. However, the decreasing trend in total reducing sugars content aligns with the pattern observed in the optical density, indicating a correlation between sugar consumption, bacterial growth, and xanthan gum production. This agrees with the findings of Miranda et al who studied the influence of strains and fermentation time on the production, composition, and properties of xanthan gum [14].

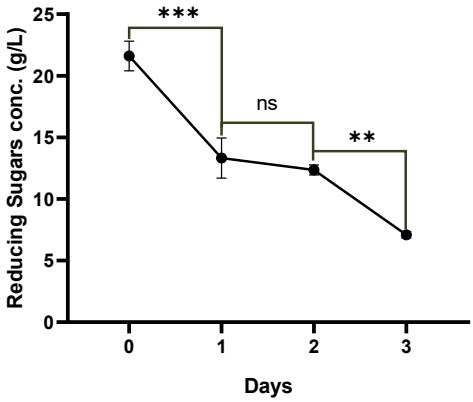

Fig. 2. Total reducing sugars content. Points are means (n= 3) and error bars are standard deviation of means, where ** is significant difference ($p < 0.0012$) and *** is significant difference ($p < 0.0001$), and ns is no significant difference ($p = 0.6821$).

3.3 Xanthan gum yield

Understanding the yield helps evaluate the efficiency of the xanthan gum production process and the productivity of the bacterial culture in converting sugars into the desired product. There is a significant ($p < 0.0001$) increase in xanthan gum yield from 3.037 g/L on Day 1 to 8.270 g/L on Day 2. This substantial rise indicates enhanced xanthan gum production between these two time points. From Day 2 to Day 3, there is a further but smaller increase in the xanthan gum yield, reaching 8.837 g/L. Although the increase is not as substantial as the previous day's change, it still indicates ongoing xanthan gum production and accumulation by the bacterial culture. The trend of increasing xanthan gum yield across the experiment indicates the continuous synthesis and accumulation of the desired product as

shown in figure 3. This increase in yield suggests that the bacteria are actively converting the available sugars from the culture medium into xanthan gum, demonstrating their metabolic activity and efficiency in producing the target biopolymer. Understanding the factors influencing this production curve, especially any limitations impacting yield increase, can help optimize the xanthan gum production process for higher efficiency and productivity.

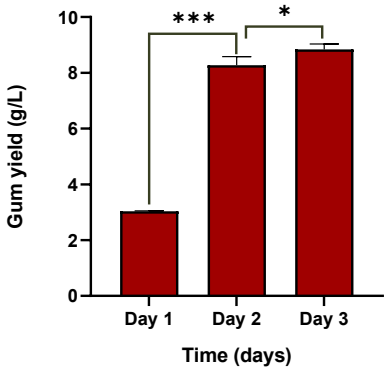

Fig. 3. Xanthan gum yield per day of biosynthesis. Bars are means (n= 3) and error bars are standard deviation of the means, where * is significant difference (p = 0.0401) and *** is significant difference (p < 0.0001).

3.4 FTIR spectra of xanthan gums

Infrared spectroscopy serves as a methodology utilized to discern similarities or disparities in the chemical structures of compounds. The xanthan gum produced underwent analysis to ascertain the functional groups within the biopolymer structure and its similarity with the commercial xanthan gum as shown in figure 4. The study encompassed spectral bands ranging from wave numbers 500 to 4000 cm^{-1}. A discernible peak within the wavenumber range of 3600 to 3000 cm^{-1} indicated the stretching vibration of hydroxyl (–OH) groups. Another shorter peak in the wavenumber range of 2900 to 2800 cm^{-1} correlated with the stretching vibration of aliphatic hydrocarbon (C–H) groups. The spectral region between 1750 and 1500 cm^{-1} denoted the stretching vibrations of C=O in pyruvate and acetyl structures. At 1249 cm^{-1}, a peak emerged, associated with the stretching of symmetric carboxylate (–COO–) in glucuronic acids. Moreover, the presence of a peak at 1022 cm^{-1} was linked to C–O–C stretching vibrations of the glycosidic bonds of xanthan gum [15]. These findings align with previous FTIR analyses of xanthan gum [15, 16], suggesting the isolated polysaccharide's spectral behavior mirrors that of the commercial gum, likely due to similarities in manufacturing techniques and analytical methodologies employed in this study. Nonetheless, relying solely on infrared spectroscopy may not conclusively determine the structure of the polysaccharides under study. Therefore, complementary techniques such as proton and carbon Nuclear Magnetic Resonance spectroscopy (NMR) are recommended to confirm the chemical structure.

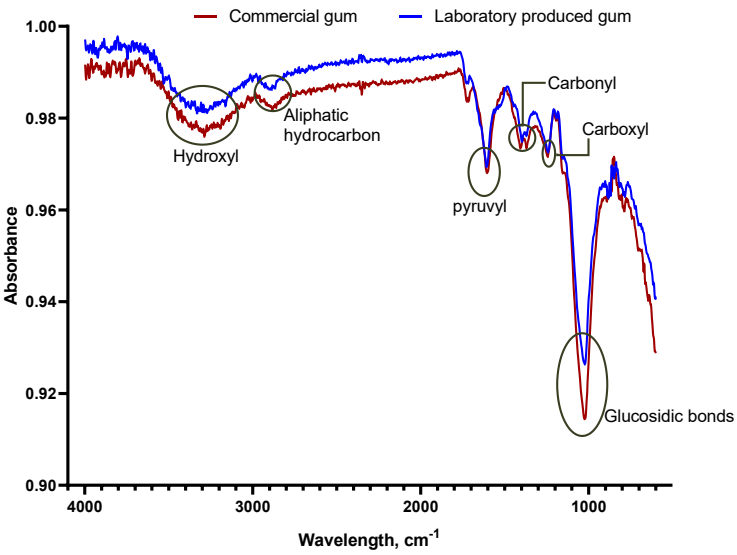

Fig. 4. FTIR spectrum of commercial and laboratory produced xanthan gum.

3.5 Effect of xanthan gum on fermented soymilk on storage

To understand the effect of the different amount of xanthan gum on the fermented soymilk, the pH changes, syneresis, water holding capacity, and lactic acid bacteria counts during a 14-day storage period were determined. The consumer acceptability of the product was also assessed.

3.5.1 pH

To guarantee the safety, quality, and consumer acceptability of the fermented soymilk it is important to monitor the pH changes. The pH may affect the microbial stability, flavor and texture, and shelf life of the product. Therefore, during the 14-day storage period, the pH of the samples was taken to ascertain the effect of xanthan on the pH changes of the fermented soymilk as represented in table 1. In this study, all the treatments including the control's pH value changed significantly ($p < 0.05$) during the 14-day storage period except for T3, whose pH value did not change significantly ($p > 0.05$) after day 7 of the storage period. This means that, the inclusion of xanthan gum up to about 0.015% could stabilize the pH of fermented soymilk compared to other inclusion levels. However, these findings agreed with that of El-Sayed et al. who obtained pH changes during a 10-day storage of soy yoghurt manufactured with laboratory-produced xanthan gum [17]. In general, the change in pH for the xanthan-supplemented samples changed to a lesser extent.

Table 1. pH changes during storage of fermented soymilk with added xanthan gum.

	Days	Control	T1	T2	T3	T4
pH	Day 1	4.69 ±0.01	4.71 ±0.01	4.74 ±0.01	4.72 ±0.01	4.73 ±0.01
	Day 7	4.62 ±0.02****	4.69 ±0.00*	4.64 ±0.01****	4.66 ±0.00****	4.68 ±0.02****
	Day 14	4.49 ±0.01**	4.62 ±0.00****	4.61 ±0.00**	4.66 ±0.00^{ns}	4.65 ±0.01***

Values are means (n=3, ±SD). Mean in the same column with * is significantly different ($p = 0.0270$), ** is significantly different ($p = 0.0028$), *** is significantly different ($p = 0.0002$), **** is significantly different ($p < 0.0001$), and ***ns*** is significantly the same ($p > 0.9999$) with respect to the preceding mean.

3.5.2 Syneresis and water holding capacity

Enhancing water holding capacity helps minimize syneresis, leading to products with better stability, improved texture, and extended shelf life [18]. In this study, the syneresis of all the samples did not change significantly ($p > 0.05$) except for T3 that changed significantly ($p < 0.05$) during the storage period. This could be as a result of the soybean variety used and the soymilk extraction method. According to Atuna et al., soybean variety could influence the soymilk composition [19] which could affect the syneresis behavior of the product. Nevertheless, from the individual values in table 2, at the end of the storage period, syneresis decreases as the xanthan gum content increases. Increasing xanthan gum contents leads to decreasing in syneresis. Regarding the WHC of the various samples, similar trends were observed. The WHC of all the samples changed significantly ($p < 0.05$) at the end of day 7 and remain significantly ($p > 0.05$) unchanged at the end of the storage period as shown in table 2. The syneresis and WHC values agrees with that of Dimitrellou et al. who no significant changes during storage period of yoghurts supplemented with juices from grapes and berries [20].

Table 2. Effect of storage on **s**yneresis and water holding capacity of fermented soymilk.

	Days	**Control**	**T1**	**T2**	**T3**	**T4**
Syneresis, %	Day 1	23.63 ±1.83	23.20 ±0.51	26.31 ±0.33	23.43 ±0.44	25.45 ±0.54
	Day 7	26.46 ±0.72^{ns}	27.75 ±0.97**	27.19 ±0.19^{ns}	27.79 ±0.63**	25.35 ±0.07^{ns}
	Day 14	27.88 ±1.14^{ns}	26.46 ±0.14^{ns}	25.01 ±1.50^{ns}	24.10 ±2.74*	24.37 ±2.13^{ns}
WHC, %	Day 1	63.76 ±6.76	55.51 ±6.43	67.18 ±5.82	66.68 ±0.11	63.79 ±5.48
	Day 7	56.25 ±0.07*	53.07 ±3.00*	59.01 ±0.60*	61.16 ±0.81*	60.52 ±0.22*
	Day 14	55.59 ±0.22^{ns}	53.41 ±2.18^{ns}	53.76 ±3.28^{ns}	57.62 ±2.52^{ns}	55.39 ±0.83^{ns}

Values are means (n=2, ±SD). Means in the same column with * are significantly different ($p < 0.05$) and means in the same column with ns are significantly the same ($p > 0.05$) with respect to the preceding mean.

3.5.3 Lactic acid bacteria count

Maintaining adequate LAB counts is necessary to ensures the probiotic during storage. A stable LAB counts in fermented soymilk during storage also ensures product safety and consistency [21]. As shown in table 3, the LAB counts in samples changed significantly ($p < 0.05$) during the storage period except for T3 and T4 whose LAB counts changed significantly ($p < 0.05$) at the end of day 7 and remain significantly the same ($p > 0.05$) at end of the storage period. However, T4 LAB counts did not change significantly ($p > 0.05$) during the whole storage period. These results suggest that increasing in xanthan content helps to maintain the LAB concentration during storage. Nevertheless, the LAB counts obtained in this study range from 8.965 ±0.04 to 9.481 ±0.02 logCFU/ml for the treated samples which is similar to that obtained by Huo et al. in fermented soymilk with various LAB [22].

Table 3. LAB counts during storage of fermented soymilk with added xanthan gum.

	Days	**Control**	**T1**	**T2**	**T3**	**T4**
logCFU /ml	Day 1	9.632 ±0.02	9.347 ±0.03	9.282 ±0.02	9.481 ±0.02	9.365 ±0.14
	Day 7	9.307 ±0.03****	8.965 ±0.04****	9.012 ±0.01****	9.148 ±0.01****	9.329 ±0.01^{ns}
	Day 14	9.180 ±0.04*	9.107 ±0.04*	9.409 ±0.03****	9.212 ±0.02^{ns}	9.332 ±0.03^{ns}

Values are means (n=2, ±SD). Means in the same column with * are significantly different ($p < 0.05$) and means in the same column with ns are significantly the same ($p > 0.05$) with respect to the preceding mean.

3.5.4 Effect of xanthan gum on consumer acceptability of fermented soymilk

Assessing the consumer acceptability of food products allows producers to make informed decisions to maintain product quality, meet consumer preferences, and ensure market success. Therefore, the effect of xanthan gum on the fermented soymilk was assessed. As shown in table 4, the organoleptic attributes studied in the different treatments did not differ significantly ($p > 0.05$) from the control. However, according to the individual scores of the overall acceptability, the treated samples are more preferred by consumers. The high score for flavor in the control samples showed that the presence of the gum could have reduce the effect on the flavor the product. This may be due to the sorption capacity of gum and a decrease in the concentration of volatile aromatic substances.

Table 4. Effect of xanthan gum on consumer acceptability of fermented soymilk.

Samples	Flavor	Appearance	Taste	Overall acceptability
Control	4.7	3.9	3.6	3.6
T1	4.2 ^{ns}	3.9 ^{ns}	3.6 ^{ns}	3.7 ^{ns}
T2	3.5 ^{ns}	3.8 ^{ns}	3.3 ^{ns}	3.5 ^{ns}
T3	3.5 ^{ns}	3.6 ^{ns}	3.3 ^{ns}	3.5 ^{ns}
T4	3.8 ^{ns}	3.9 ^{ns}	3.6 ^{ns}	3.7 ^{ns}

Values are means, where ***ns*** = no significant difference ($p > 0.05$). 1-dislike extremely, 2-dislike, 3-neither like nor dislike, 4-like, 5-like extremely.

4 Conclusion

This comprehensive investigation encompassed both the biotechnological aspect of xanthan gum production and its application in fermented soymilk, aiming to contribute valuable insights to the fields of food science, biotechnology, and industrial applications.

From this work, the xanthan gum produced with maximum yield of 8.837 g/L. The gum is in conformity of the commercial gum according to the FTIR spectrum and has stabilized the probiotics content of the product on storage. Addition of the gum has also helped to fairly maintain pH and syneresis during storage, even in such a small quantity (0.005% - 0.020%). In general, the formulated product has the tendency to have extended shelf-life (more than 14 days) based on the results obtained. Also, the health benefits including the antioxidant activity that xanthan naturally possessed could be additional product added to this formulated product.

The study's findings provided a foundation for harnessing the potential of xanthan gum in enhancing the quality and marketability of fermented soymilk, further expanding its utility in the ever-evolving landscape of functional food products. In addition, the optimization of the production parameters for xanthan gum, and further characterization and the rheological behavior of the gum produced and formulated fermented soymilk, are considered as the limitations of this study, and must be considered in further studies. The effect on the aroma of the product when adding xanthan must be taken into account when developing new products.

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